

FIG. I

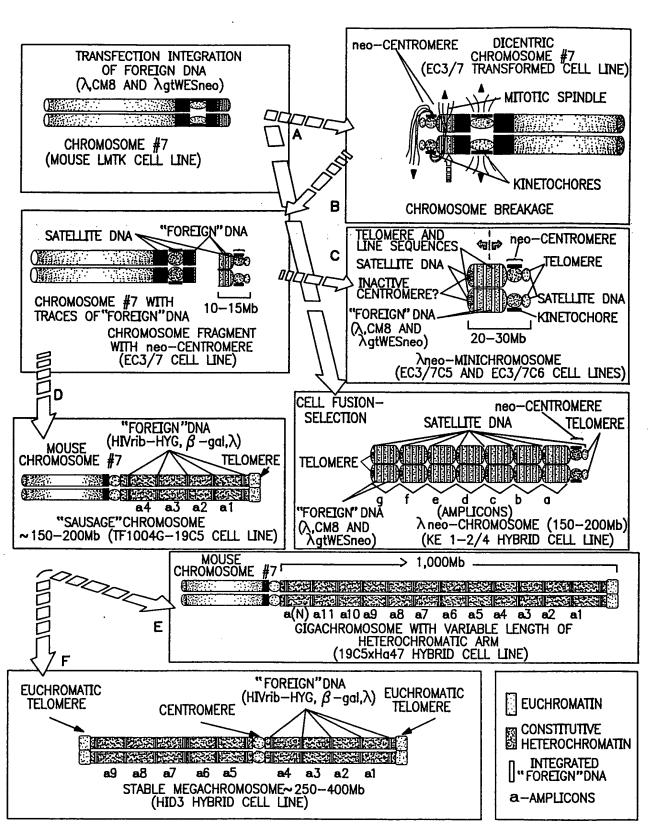
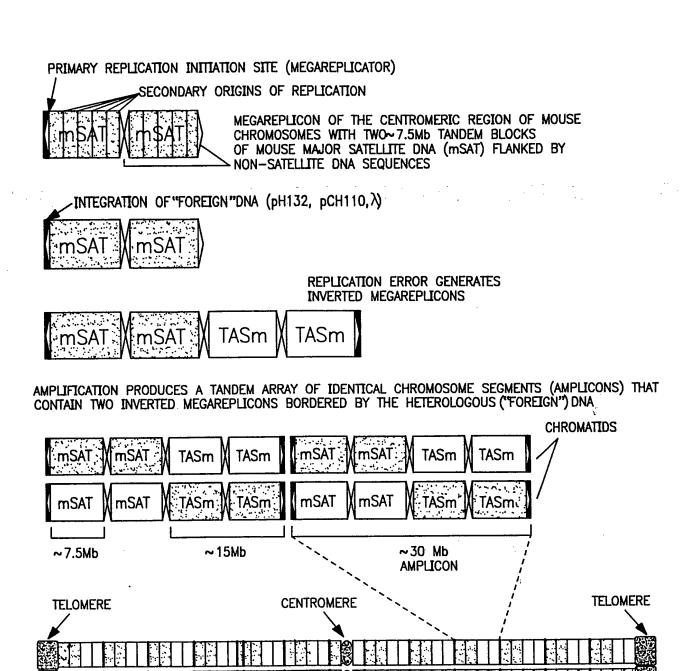


FIG. 2



STABLE MEGACHROMOSOME (~250-400Mb)

FIG. 3

Figure 4

EC3/7 Mouse LMTK fibroblast cell line with neo-centromere

(EADLACKY STAL Proc. Natl. Acad. Sci. USA, 88: 3106-3110, 1991)

Deposited in the European Collection of Animal Cell Culture (ECACC)

Single-cell subcloning

EC3/7C5 Mouse LMTK fibroblast cell lines with neo-minichromosome
(HADIACZKY LT AL. Proc. Natl. Acad. Sci. USA., 88: 8106-8110, 1991)

Cotransfection with plasmids pH132 (HTVribazyme, hygromycin resistance) pCH110 (\$\beta_{\text{galactosidase}}\$), and lambda phage (\$\lambda cl 875 Sam7) DNA, selection with Hygromycin B.

TF1004G-19C5* - Mouse LMTK fibroblast cell lines with neo-minichromosome, and stable "sausage" chromosome

Fusion with Chinese hamster (CHO ICO) cell line, selection with Hygromycin B and HAT.

19C5xH24 - Mouse-hamster hybrid cell line carrying the neo-minichromosome and the "sausage" chromosome, containing complete hamster genome and partial mouse genome.

Brill treatment, single cell cloning, selection: G418 (Neomycin) or Hygromycin, or hoth

G3D5* - Mouse-hamster hybrid call line carrying the neo-minichromosome and the megachromosome, containing complete hamster genome and partial mouse genome.

H1D3* - Mouse humster hybrid cell line carrying no neo-minichromosome but the megachromosome is present, containing complete hamster genome and partial mouse genome.

Fusion with CD4+ HcLa cell line carrying the CD41 and Neomycin resistance gene plasmid construct (CD4neo), selection with C418 and Tygromycin B

HixHe41* - Mouse-hamster-human hybrid cell line carrying the megachromosome present, containing complete hamster genome, and partial mouse genome, and a single human chromosome with integrated CD4nco construct (UNIVIDELISIED).

Repeated 3rdU treatment, single-cell climing

1B3 - Same as H1xHe41, but approximately 25% of the cells are carrying a truncated megacinomosome

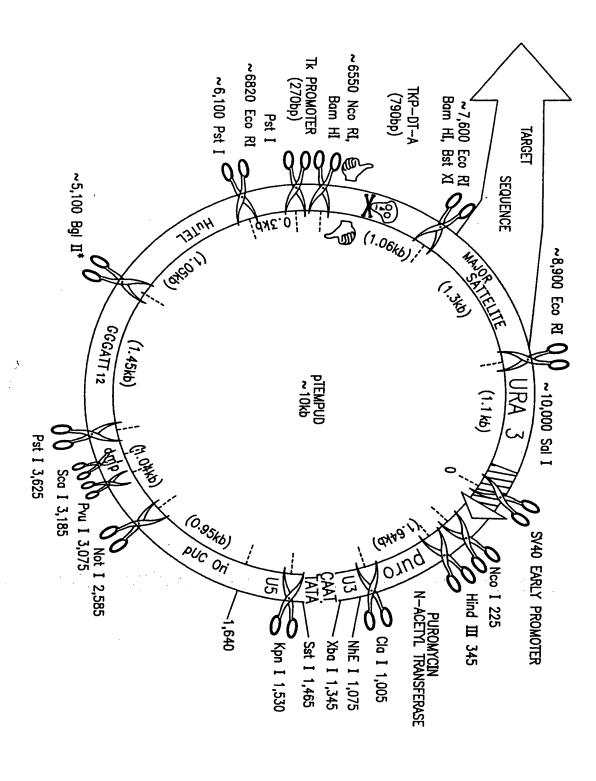


FIG. 5